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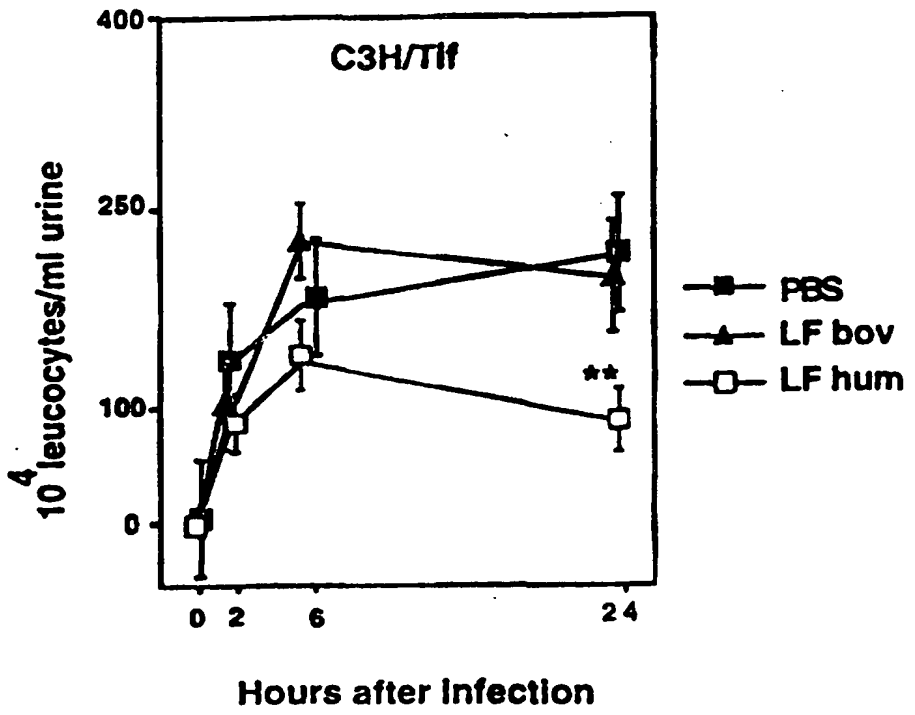
(54) Title: TREATMENT AND PREVENTION OF INFECTIONS, INFLAMMATIONS AND/OR TUMOURS WITH LACTOFERR AND/OR LACTOFERRICIN

(57) Abstract

The present invention relates to a pharmaceutical composition comprising lactoferrin and/or lactoferricin for treatment and/or prevention of infections, inflammations and/or tumours, to the use of lactoferrin and lactoferricin in the production of a pharmaceutical composition for treatment and/or prevention of infections, inflammmations and tumours, and to a method for treatment and/or prevention of infections, inflammations and/or tumours comprising administration of lactoferrin and/or lactoferricin. The invention is particularly well suited for treatment and/or prevention of urinary tract infections and colitis. The lactoferrin and/or lactoferricin according to the present invention is preferably orally administered. Furthermore, the composition comprising lactoferrin and/or lactoferricin may be included in an infant formula food.



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TREATMENT AND PREVENTION OF INFECTIONS, INFLAMMATIONS AND/OR TUMOURS WITH LACTOFERRIN AND/OR LACTOFERRICIN

FIELD OF THE INVENTION

The present invention relates to a pharmaceutical composition comprising lactoferrin and/or lactoferricin for treatment and/or prevention of infections, inflammations and/or tumours, to the use of lactoferrin and lactoferricin in the production of a pharmaceutical composition for treatment and/or prevention of infections, inflammations and tumours, and to a method for treatment and/or prevention of infections, inflammations and/or tumours comprising administration of lactoferrin and/or lactoferricin.

BACKGROUND OF THE INVENTION

It is known that human milk in several ways is anti-inflammatory. Goldman et al. pointed out that human milk is poor in initiators and mediators of inflammation but rich in anti-inflammatory agents (see Goldman A. S., et al., Anti-inflammatory properties of human milk, Acta Paediatr. Scand. 75:689-695, 1986). Human milk contains several soluble anti-infective components, such as specific secretory IgA (SIgA) antibodies and non-specific components, including lactoferrin (LF) (see e.g. Hanson L. Å., et al., Protective factors in milk and the development of the immune system, Pediatrics 75:172-176, 1983).

Lactoferrin is a single chain metalbinding glycoprotein with a molecular weight of 77 kd. It occurs in three isoforms: LF-α, LF-β, and LF-γ. These three variants have the same physical, chemical and antigenic characteristics, but differ in their functional properties.

The iron-binding lactoferrin is also present in specific granules of polymorphonuclear leucocytes and in other exocrine secretions than milk such as saliva, tears and bronchial mucus, as well as cervical secretion, amniotic fluid, decidua, and trophoblasts (see e.g. Montreuil

J., et al., Isolement d'une lactosiderophiline du lait de femme, CR Acad. Sci. Paris 250 D:1736-37, 1960; Montreuil J., et al., Preparation et propriétés de la lactosiderophiline (lactotransferrine) du fait de femme, Biochim. Biophys. Acta 45:413-421, 1960; and Masson P. L., et al., Lactoferrin an ironbinding protein neutrophilic leucocytes, J. Exp. Med. 130:643-656, 1969). Lactoferrin is associated with host defense at mucosal surfaces through its antibacterial and iron-binding properties.

Human lactoferrin is found in colostrum and mature milk at levels of 2-5 g/l.

Bovine lactoferrin shares 68% and 64% amino acid identity with human lactoferrin and murine lactoferrin, respectively.

Lactoferricin is a pepsin-cleaved fragment of human and bovine lactoferrin. It has recently been found to contain the structural domain responsible for the bactericidal properties of lactoferrin (see e.g. Bellamy W., et al., Identification of the bactericidal domain of lactoferrin, Biochim. Biophys. Acta 1121:130-136, 1992, and Bellamy W., et al., Antibacterial spectrum of lactoferricin B, a potent bactericidal peptide derived from the N-terminal region of bovine lactoferrin, J. Appl. Bact. 73:472-479, 1992).

Lactoferrin receptors are found on many types of cells including monocytes and macrophages (Broxmeyer H. E., et al., Specificity and modulation of the action of lactoferrin, a negative feedback regulator of myelopoiesis, Blood 55:324-333, 1980), lectin-stimulated human peripheral blood lymphocytes (Mazurier J., et al., Expression of human lactotransferrin receptors in phytohemagglutinin-stimulated human peripheral blood lymphocytes. Isolation of the receptors by anti-ligand-affinity chromatography, Eur. J. Biochem. 179:481-487, 1989), brush-border cells (Hu W. L., et al., Lactotransferrin receptor of mouse small-intestinal brush border. Binding characteristics of membrane-bound and Triton X-100-

solubilized forms, Biochem. J. 249:435-441, 1988; Cox T. M., et al., Iron-binding proteins and influx of iron across the duodenal brush border. Evidence for specific lactotransferrin receptors in the human intestine. Biochim. Biophys. Acta 588:120-128, 1979; Mazurier J., et al., Visualization of lactotransferrin brush-border receptors by ligand-blotting. Biochim. Biophys. Acta 821:453-460, 1985; and Wei-Lu Hu, et al., Isolation and partial characterization of a lactotransferrin receptor from mouse intestinal brush-border, Biochemistry 29:535-540, 1990), tumor cell lines, e.g. HT-29, HL-60, K562, (see e.g. Roiron D., et al., Lactoferrin-binding sites at the surface of HT29-D4 cells. Comparison with transferrin. Eur. J. Biochem. 186:367-373, 1989; Miyazawa K., et al., Effect on lactoferrin binding to monocyte/macrophage-differentiated HL-60 cells. J Immunol. 146:723-729, 1991; and Yamada Y., et al., Lactoferrin binding by leukemia cell lines, Blood 70:264-270, 1987).

In addition to the role of lactoferrin as an essential growth factor for both human B- and T-lymphocytic cell lines (see e.g. Hashizume S., et al., Identification of lactoferrin as an essential growth factor for human lymphocytic cell lines in serum-free medium, Biochem. Biophys. Acta 763:377-382, 1983) and as an inducer of growth of HT-29 cells (see e.g. Anuric M., et al., Effect of lactoferrin on the growth of a human colon adenocarcinoma cell line - comparison with transferrin. In Vitro 20:543-548, 1984), lactoferrin is a negative regulator of myelopoiesis (see e.g. Broxmeyer H. E., et al., Specificity and modulation of the action of lactoferrin, a negative feedback regulator of myelopoiesis, Blood 55:324-333, 1980, and Gentile P., et al., Suppression of mouse myelopoesis by administration of human lactoferrin in vivo and the comparative action of human transferrin, Blood 61:982-993, 1983). This latter function is mediated through suppression of IL-1 and GM-CSF release from monocytes and macrophages (see e.g. Broxmeyer H. E., et al.,

Lactoferrin acts on I-A and I-E/C antigen subpopulations of mouse peritoneal macrophages in the absence of T lymphocytes and other cell types to inhibit production of granulocyte-macrophage colony stimulatory factors "in vitro", J. Immunol. 133:306-314, 1984, and Zucali J. R., et al., Lactoferrin decreases monocyte-induced fibroblast production of myeloid colony-stimulating activity by suppressing monocyte release of interleukin-1, Blood 74:1531-1536, 1989).

After binding of bacterial lipopolysaccharides (LPS) to macrophages, T-cells and cultured human monocytes, these cells synthesize tumor necrosis factor-α (TNF-α), interleukin-1 (IL-1), interleukin-6 (IL-6), and colony-stimulating factor (CSF) (see e.g. Arai K., et al., Cytokines: coordinators of immune and inflammatory responses, Ann. Rev. Biochem. 59:783-836, 1990; Hirano T., et al., Biological and clinical aspects of interleukin 6, Immunol. Today 11:443-449, 1990; and Shalaby M. R., et al., Endotoxin, tumor necrosis factor-α and interleukin-1 induce interleukin-6 production "in vivo", Clin. Immunol. Immunopath. 53:488-498, 1989). Cells participating in the inflammatory response carry several different LPS-binding receptors (Lei M-G., et al., Specific endotoxic lipopolysaccharide-binding proteins on murine splenocytes. II. Membrane localization and binding characteristics, J. Immunol. 141:1006-1011, 1988 and Couturier C., et al., Binding sites for endotoxin (LPS) on human monocytes, J. Immunol. 147:1899-1904, 1991). Such cells also have receptors for lactoferrin. An interaction between LPS and lactoferrin has been observed, the complex being bound to the cells also via LPS receptors (see Miyazawa K., et al., Effect on lactoferrin binding to monocyte/macrophage-differentiated HL-60 cells. J Immunol. 146:723-729, 1991). Recently, it was showed that lactoferrin exerted an inhibitory effect on the production of IL-1 and TNF-α in LPS stimulated monocytes (see Crouch P. M., et al., Regulation of cytokine release from mononuclear cells by

the iron-binding protein lactoferrin, Blood 80:235-240, 1992).

It has earlier been shown (see Mattsby-Baltzer I. et al., Lactoferrin or a fragment thereof inhibits the endotoxin-induced interleukin-6 response in human monocytic cells, Pediactric Research 40:257-262, 1996) that human and bovine lactoferrin as well as bovine lactoferricin suppress LPS-induced IL-6 response when added to fresh monocytes or cultured monocytic cells. Human lactoferrin has also been reported to suppress TNF-α induced IL-6 response when added to fresh monocytes or cultured monocytic cells.

According to the present invention it has now been found that lactoferrin and lactoferricin have an in vivo effect on all kinds of inflammation, i.e. not only when IL-6 is involved, as well as on infections, such as urinary tract infection, and tumours.

DESCRIPTION OF THE INVENTION

Thus, an object of the present invention is to provide a pharmaceutical composition for treatment and/or prevention of infections, inflammations and/or tumours comprising an effective amount of lactoferrin and/or lactoferricin.

Another object of the present invention is use of lactoferrin and/or lactoferricin in the production of a pharmaceutical composition for treatment and/or prevention of infections, inflammations and/or tumours.

A third object of the present invention is to provide a method for treatment and/or prevention of infections, inflammations and/or tumours by administration of an effective amount of lactoferrin and/or lactoferricin.

The characterising features of the invention will be evident from the following description and the appended claims.

In order to treat a patient, suffering from an infection, an inflammation or a tumour, with the pharmaceu-

tical composition according to the invention, the pharmaceutical composition, comprising an effective amount of lactoferrin and/or lactoferricin, is preferably administered systemically, and most preferably orally.

The infections treatable with the pharmaceutical composition according to the present inventions include infections caused by all kinds of pathogens, such as bacteria, viruses, fungi, etc.

Inflammation is a phenomenon marked by abnormal "redness" and swelling of tissues and organs, pain and heat in affected areas, capillary dilation, leucocyte infiltration, etc. Inflammation is primarily caused by exposure to bacterial and other noxious agents and physical injury. Inflammation is mediated by a variety of cytokines and other chemical signals. These mediators of inflammation include tumor necrosis factor-α (TNF-α), interleukin-1 (IL-1), interleukin-6 (IL-6), interleukin-8 (IL-8), and various colony-stimulating factors (CSFs).

As used herein, "treatment" refers to preventing, curing, reversing, attenuating, alleviating, minimizing, suppressing or halting the deleterious effects of a disease state, disease progression or other abnormal condition, including urinary tract infections.

"Prevention" refers to minimizing, reducing or suppressing the risk of developing a disease state or progression or other abnormal or deleterious conditions.

A "patient" is a subject at risk for or suffering from a disease state, disease progression or other abnormal or deleterious condition.

An "effective amount" is an amount sufficient to treat or prevent a disease state, disease progression or other abnormal or deleterious condition.

"Systemic administration" can be undertaken by oral, nasal, intravenous, intraartery, intracavitary, intramuscular, subcutaneous, transdermal, suppositories (including rectal) or other routes known to those of skill in the art. Preferably, the pharmaceutical composi-

tion according to the present invention is formulated for oral administration.

The lactoferrin and lactoferricin used according to the present invention can e.g. be obtained through isolation and purification from natural sources, such as human milk, through use of genetic engineering techniques, such as recombinant expression or direct production in genetically altered animals, or through chemical synthesis. The lactoferricin can also be obtained by enzymatic degradation of lactoferrin (hydrolysate).

The lactoferrin used according to the present invention is preferably human lactoferrin or bovine lactoferrin, and it is preferably administered as a hydrolysate.

The lactoferricin used according to the present invention is preferably human lactoferricin or bovine lactoferricin.

The pharmaceutical composition comprising lactoferrin and/or lactoferricin according to the present invention is particularly well suited for treatment and/or prevention of urinary tract infection and colitis, but several other inflammatory and infectious diseases are also treatable according to the present invention, such as inflammatory bowel diseases, rheumatoid arthritis, conditions caused by the virus HIV-1, conditions caused by the virus CMV, and conditions caused by the fungus Candida albicans.

The pharmaceutical composition according to the present invention is also well suited for preventive medical care by reducing the risk of developing urinary tract infection or other inflammatory or infectious diseases in patients with an increased risk of attracting such complications.

The pharmaceutical composition according to the present invention may also comprise other components, such as pharmaceutically acceptable carriers, vehicles, preservatives, lubricators etc., which is well known to persons skilled in the art.

According to the present invention it is also possible to include lactoferrin and/or lactoferricin, in an effective amount, in any kind of food or beverage intended to reduce infections and/or inflammations in patients running an increased risk of such conditions due to an underlying disease or a medical treatment.

According to the present invention it is also possible to include lactoferrin and/or lactoferricin, in an effective amount, in an infant formula food intended to inhibit harmful effects of bacteria, such as weight loss caused by inflammation induced by bacteria, viruses or fungi in infants.

EXAMPLES

The invention will now be further explained in the following examples. These examples are only intended to illustrate the invention and should in no way be considered to limit the scope of the invention.

In the examples reference is made to the accompanying drawings on which:

Fig. 1 a - d illustrate bacterial recovery from the kidney (a and b) and bladder (c and d), respectively, of C3H/Tif and C3H/HeN mice infected with E. coli in the urinary tract and perorally given human lactoferrin (LF hum), bovine lactoferrin (LF bov), or PBS, 30 min after the injection of bacteria. The samples represented by symbols below the line were culture negative.

Fig. 2 a and b illustrate the kinetics of the urinary leucocyte influx in E. coli infected C3H/Tif and C3H/HeN mice treated with human lactoferrin (LF hum), bovine lactoferrin (LF bov), or PBS.

Fig. 3 a and b illustrate the kinetics of the urinary IL-6 response in E. coli infected C3H/Tif and C3H/HeN mice treated with human lactoferrin (LF hum), bovine lactoferrin (LF bov), or PBS, 30 min after the injection of bacteria.

Fig. 4 illustrates the serum IL-6 response 24 h after experimentally induced urinary tract infection in C3H/Tif and C3H/HeN mice treated with human lactoferrin (LF hum), bovine lactoferrin (LF bov), or PBS, 30 min after the injection of bacteria.

Fig. 5 illustrates the cytokine concentration in serum from mice with experimentally induced colitis after treatment with bovine lactoferrin (LF bov) compared to a control group not receiving lactoferrin.

Example 1: Treatment of urinary tract infection in mice by oral administration of human lactoferrin

The antibacterial and anti-inflammatory properties of lactoferrin were explored by studying the effects of lactoferrin given to mice (C3H/Tif and C3H/HeN) with experimentally induced urinary tract infection (UTI).

In order to induce urinary tract infection (UTI) in the mice, the animals were injected with 100 µl of a bacterial solution containing $2x10^9$ E. coli-bacteria/ml diluted with phosphate-buffered saline (PBS) directly into the bladder via a catheter according to Svanborg-Edén et al (see C. Svanborg-Edén et al Infect. Immun. 55:1224-1232, 1987).

A solution containing 10 mg/ml of either human lactoferrin, bovine lactoferrin, or bovine lactoferricin was orally administered (50 µl) to the mice 30 min after the instillation of bacteria.

Urine samples from the mice were collected 0, 2, 5, and 24 hours after infection. 50 µl of each of the undiluted urine samples were cultured. The number of leucocytes in uncentrifuged urine was analyzed for each sample. The remaining urine from each animal at each sampling time was centrifuged and saved for IL-6 analysis.

After 24 h the mice were bled and killed. The bladder and kidneys were taken out aseptically. The organs were homogenized, and serial dilutions thereof (bladder

1/1, 1/10, kidneys 1/1, 1/10, 1/100, 1/1000) were cultured on Drigalsky plates.

The results are illustrated below in Table 1 and in Figures 1-4.

Table 1

	p values[1] from statistical comparison between LF treatment, LFcin treatment and no treatment			
	human LF	bovine LF		LFcin
mouse strain:	Tif	Tif	HeN	Tif
	n=15	n=14	n=18	n=5
Bacteria (CFU[2]) in:				
bladder	<0.0001	0.0061	0.0017	0.001
kidney	0.0057	0.0241	0.0410	0.0020
Leucocytes in the urine after:				
2 h	no	no	no	no
5 h	no	no	no	no
24 h	0.0066 (↓)	no	no	[0.0317 (↓)][4]
IL-6 in the urine after:				
2 h	[0.043 (↓)][4]	no	0.012 (↓)	no
5 h	no	[0.0418 (↑)][4]	no	no
24 h	no	no	[0.0386 (↓)][4]	no
IL-6 in serum after 24 h	0.0185 (↓)	no	0.0095 (↓)	no

[1] p values, Mann-Whitney

[2] CFU = colony forming units

[3] no = not statistically significant

[4] when four comparisons are made with one group, a significant p value should be adjusted to $p<0.025$

↑ illustrates a significant increase in the treatment group compared to the infected but untreated control group.

↓ illustrates a significant decrease in the treatment group compared to the infected but untreated control group.

The data shown in Table 1 clearly shows the effect of the treatment with lactoferrin and lactoferricin.

From the table and the figures it is evident that orally administered lactoferrin (both human and bovine) significantly decreased the number of bacteria in the urinary tract of the infected mice, compared to the control group.

In Figures 2 a and b the kinetics of the urinary leucocyte influx is illustrated (** in Figure 2 a signifies $p<0.01$, Mann-Whitney test), and in Figure 3 a and b the IL-6 response in urine is illustrated (* in these figures signifies $p<0.05$, Mann-Whitney test). These figures clearly shows that the local inflammatory response was reduced after 24 h.

The systemic cytokine response, viz. IL-6 response in serum, after 24 h is illustrated in Figure 4, and this response was also reduced in the lactoferrin treated animals.

In conclusion these results demonstrate that oral administration of lactoferrin or lactoferricin is systemically effective by preventing infection and inflammation in the urinary tract by an as yet unidentified mechanism.

Example 2: Treatment of experimental colitis by oral administration of human lactoferrin

Acute colitis was induced in C57BI/6J mice by giving 5% dextransulphate in the drinking water for 6 days. Human lactoferrin was orally given to ten mice twice a day in a dose of 1 mg/mouse, starting from day 3 of the experiment. Two control groups (in total 17 mice) were given the same volume of drinking water or bovine serum albumin (BSA) (2 mg per mouse and day). 30% of the mice in the lactoferrin treated group presented gross rectal bleeding on day 5 and 6 compared to 100% in the control group ($p = 0.0007$, Fischer's test). Moreover, the colon length was significantly reduced in the control groups

compared with the lactoferrin treated group, indicating a more advanced inflammation of the colon tissue in the controls ($p = 0.041$, Mann-Whitney test). High concentrations of lactoferrin were found in serum of the lactoferrin treated group.

In an other experiment using 3% dextransulphate the systemic TNF-α response was reduced in the lactoferrin treated mice after 10 days ($p < 0.0006$). The result is illustrated in Figure 5.

In summary, the results demonstrate that oral administration of LF reduces some of the clinical symptoms of experimental colitis.

CLAIMS

1. A pharmaceutical composition for treatment and/or prevention of infections, inflammations and/or tumours comprising an effective amount of lactoferrin and/or lactoferricin.

2. A pharmaceutical composition according to claim 1 intended for oral administration.

3. A pharmaceutical composition according to claim 1 or claim 2, intended for treatment and/or prevention of urinary tract infection.

4. A pharmaceutical composition according to claim 1 or claim 2, intended for treatment and/or prevention of colitis.

5. A pharmaceutical composition according to any one of claims 1-4, wherein the lactoferrin and/or the lactoferricin is derived from a human or bovine source.

6. A pharmaceutical composition according to any one of claims 1-5, wherein the lactoferrin is included in the pharmaceutical composition as a hydrolysate.

7. An infant formula food comprising the pharmaceutical composition according to any one of claims 1-6.

8. Use of lactoferrin and/or lactoferricin in the production of a pharmaceutical composition for treatment and/or prevention of infections, inflammations and/or tumours.

9. Use of lactoferrin and/or lactoferricin according to claim 8, wherein the pharmaceutical composition is intended for oral administration.

10. Use of lactoferrin and/or lactoferricin according to claim 8 or claim 9, wherein the pharmaceutical composition is intended for treatment and/or prevention of urinary tract infection.

11. Use of lactoferrin and/or lactoferricin according to claim 8 or claim 9, wherein the pharmaceutical composition is intended for treatment and/or prevention of colitis.

12. Use of lactoferrin and/or lactoferricin according to any one of claims 8-11, wherein the lactoferrin and/or the lactoferricin is derived from a human or bovine source.

13. Use of lactoferrin according to any one of claims 8-12, wherein the lactoferrin is included in the pharmaceutical composition as a hydrolysate.

14. Use of lactoferrin and/or lactoferricin according to any one of claims 8-13, wherein the pharmaceutical composition constitutes or is included in an infant formula food.

15. A method for treatment and/or prevention of infections, inflammations and/or tumours whereby an effective amount of a substance chosen from the group consisting of lactoferrin and lactoferricin is administered to a patient.

16. A method according to claim 15, wherein the substance is orally administered.

17. A method according to claim 15 or claim 16, used for treatment and/or prevention of urinary tract infection.

18. A method according to claim 15 or claim 16, used for treatment and/or prevention of colitis.

19. A method according to any one of claims 15-18, wherein the lactoferrin and/or the lactoferricin is derived from a human or bovine source.

20. A method according to any one of claims 15-19, wherein the lactoferrin is used in the form of a hydrolysate.

21. A method according to any one of claims 15-20, wherein the substance is included in an infant formula food.

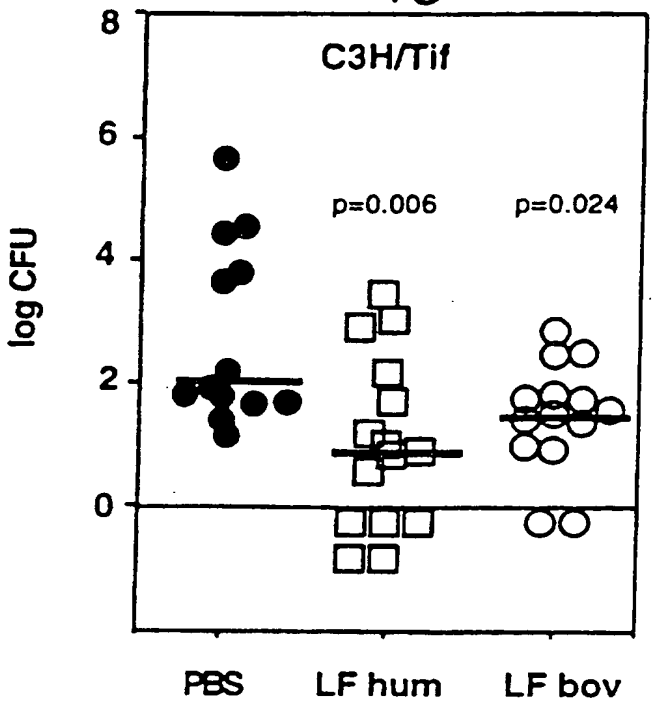
C3H/Tif
p=0.006
p=0.024
log CFU
8
6
4
2
0
PBS
LF hum
LF bov

Fig. 1a

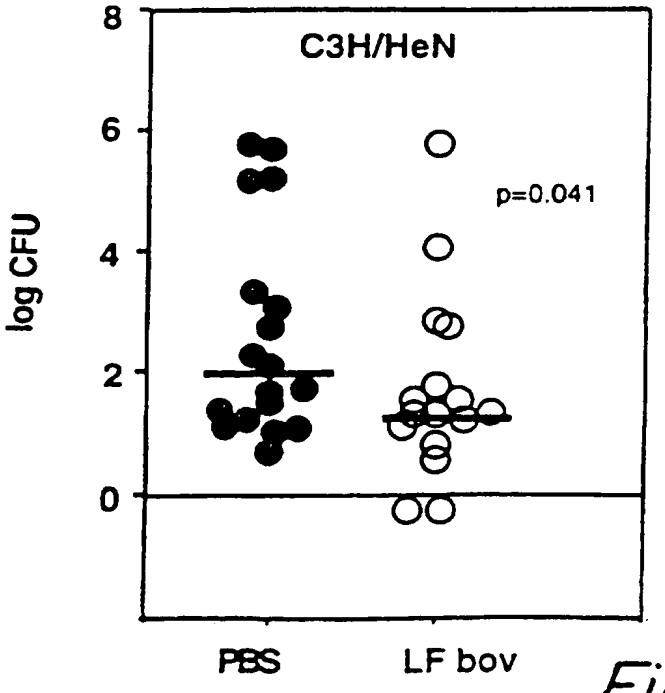
C3H/HeN
p=0.041
log CFU
8
6
4
2
0
PBS
LF bov

Fig. 1b

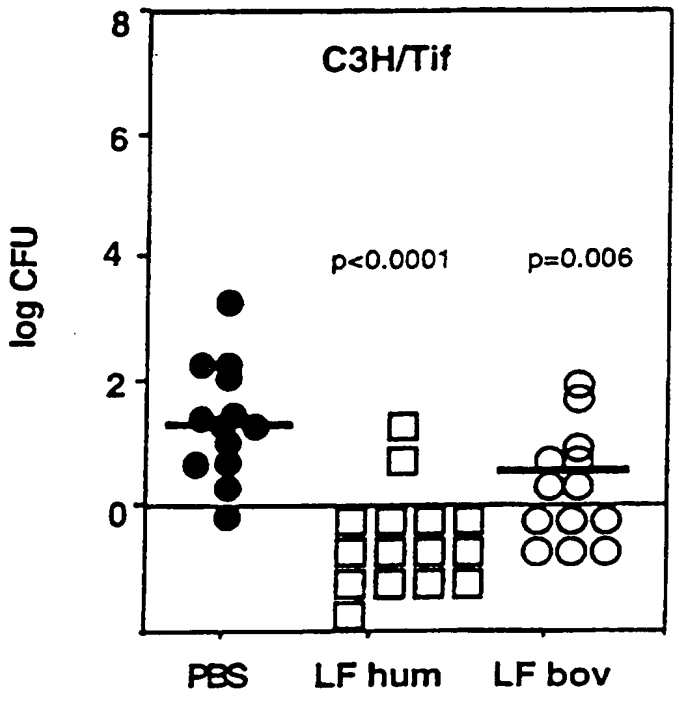
C3H/Tif
log CFU
8
6
4
2
0
p<0.0001
p=0.006
PBS
LF hum
LF bov

Fig. 1c

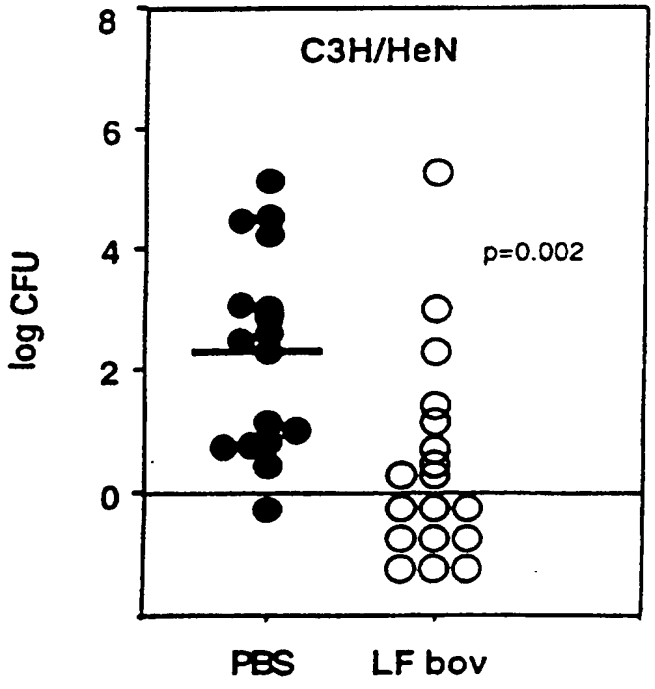
C3H/HeN
log CFU
8
6
4
2
0
p=0.002
PBS
LF bov

Fig. 1d

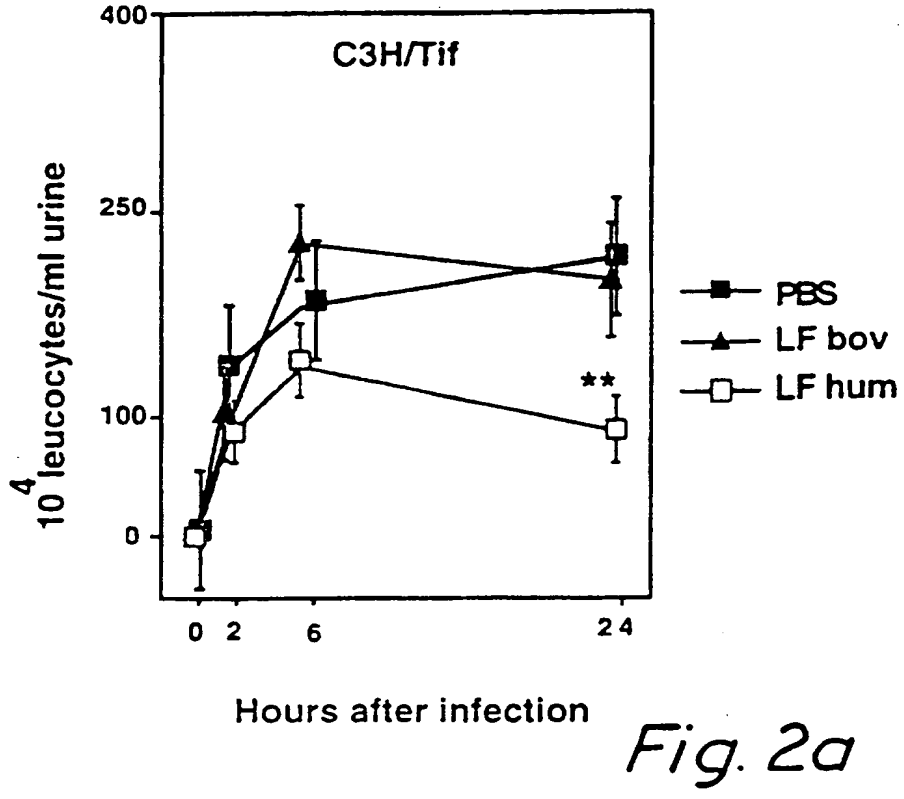
C3H/Tif
400
250
100
0
10^4 leucocytes/ml urine
**
0 2 6 24
Hours after infection
PBS
LF bov
LF hum

Fig. 2a

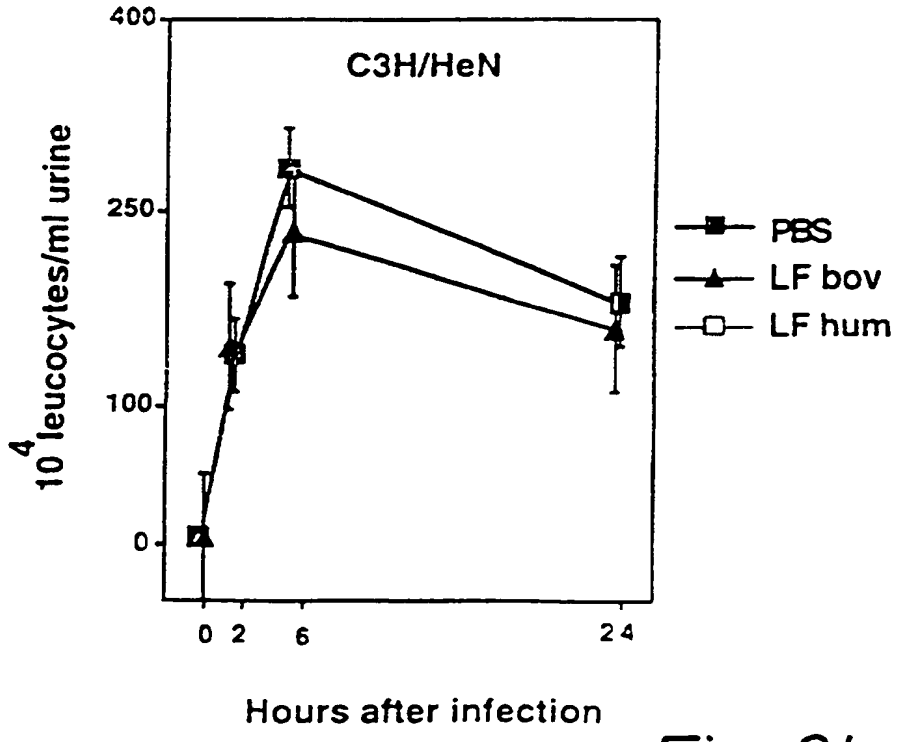
C3H/HeN
400
250
100
0
10^4 leucocytes/ml urine
0 2 6 24
Hours after infection
PBS
LF bov
LF hum

Fig. 2b

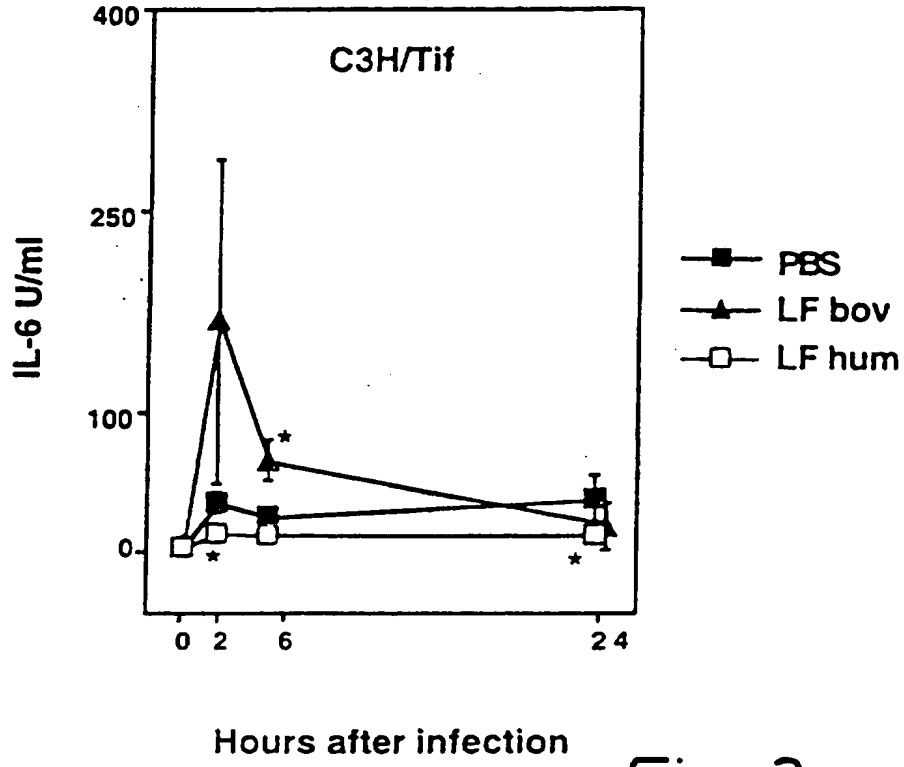
C3H/Tif
IL-6 U/ml
400
250
100
0
0 2 6 24
PBS
LF bov
LF hum
Hours after infection

Fig. 3a

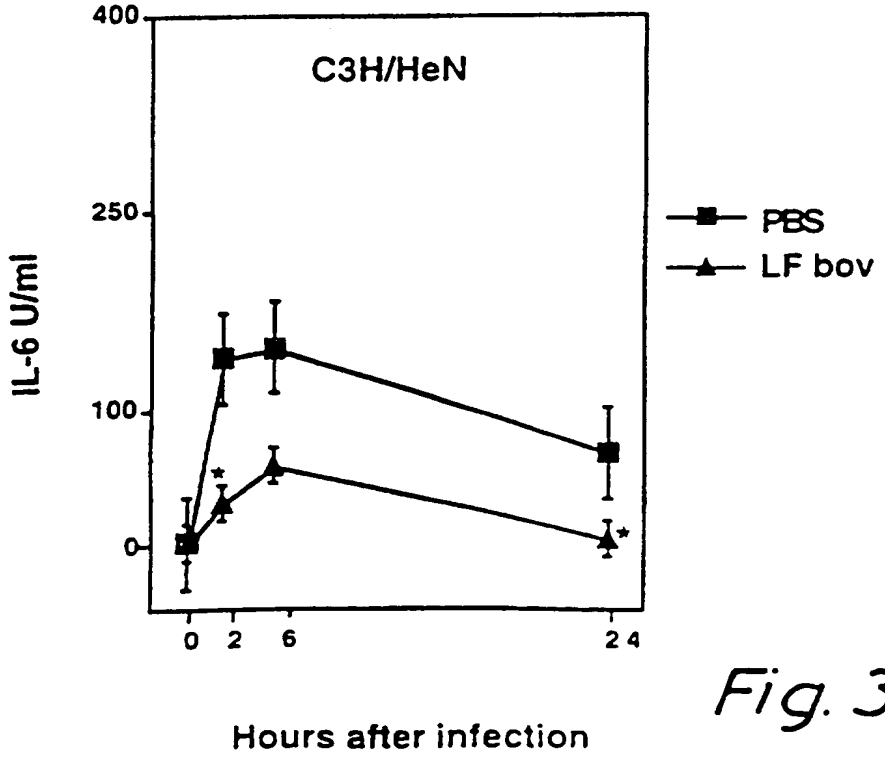
C3H/HeN
IL-6 U/ml
400
250
100
0
0 2 6 24
PBS
LF bov
Hours after infection

Fig. 3b

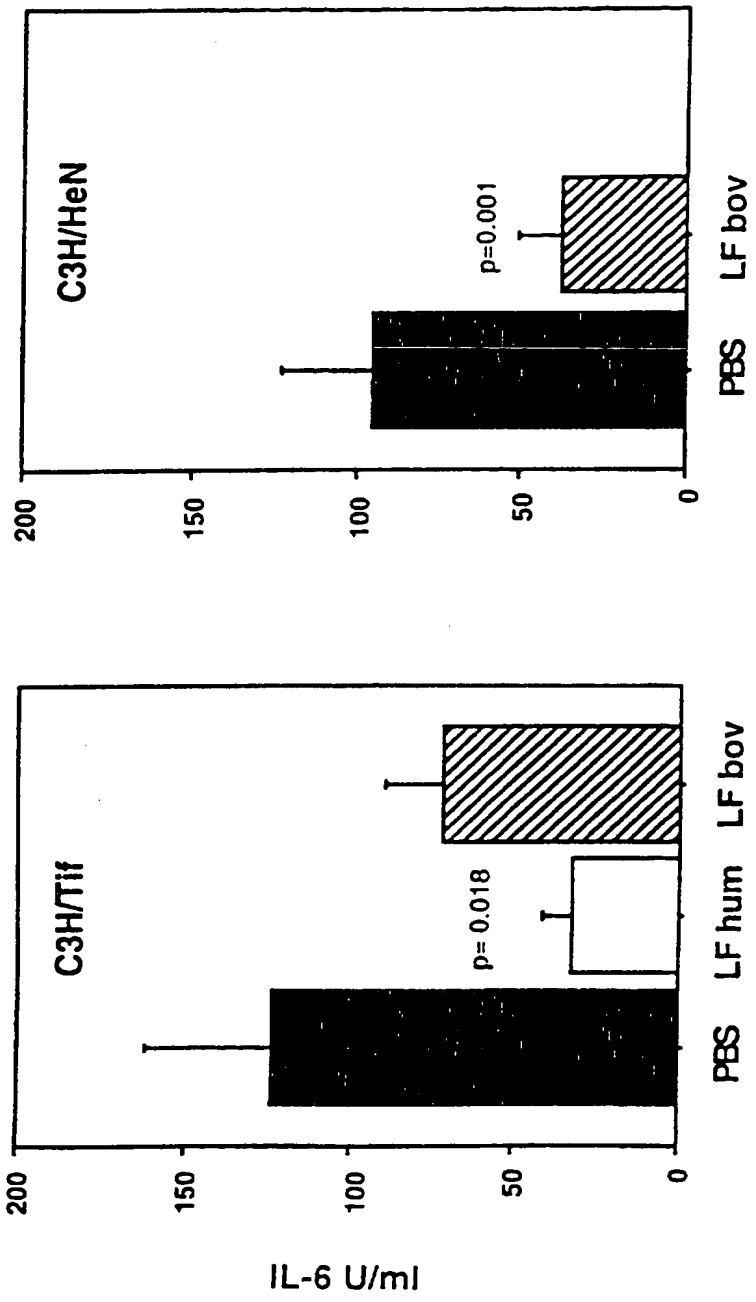
C3H/HeN
p=0.001
PBS
LF bov
200
150
100
50
0
C3H/Tif
p= 0.018
PBS
LF hum
LF bov
200
150
100
50
0
IL-6 U/ml

Fig. 4

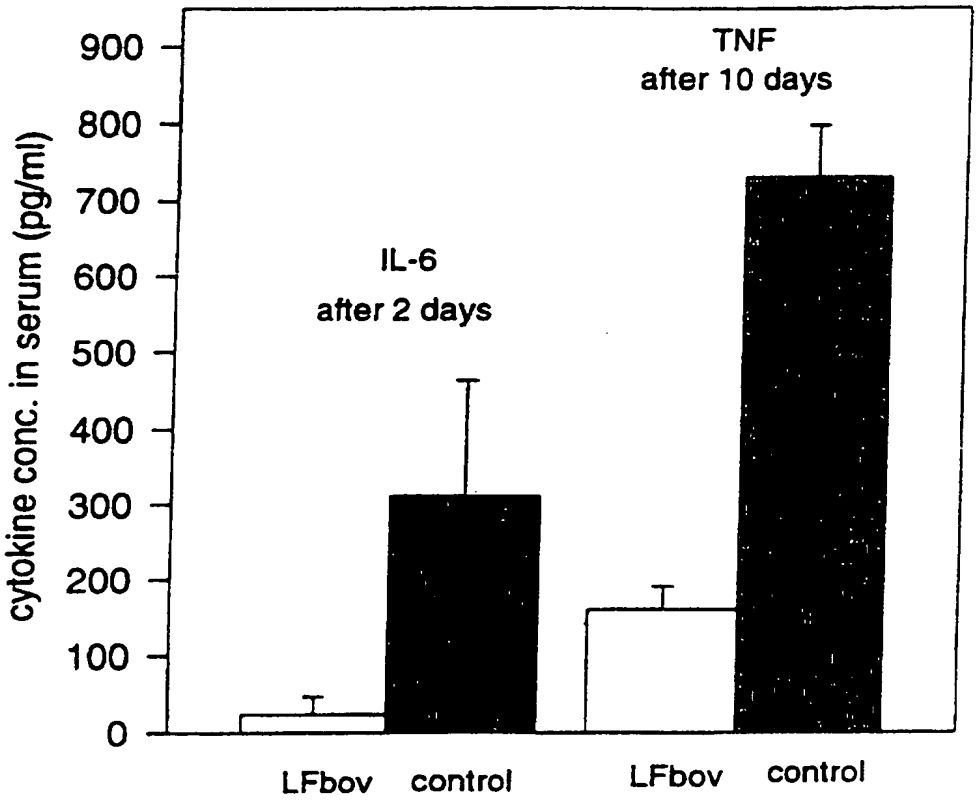
900
800
700
600
500
400
300
200
100
0
cytokine conc. in serum (pg/ml)
IL-6
after 2 days
TNF
after 10 days
LFbov
control
LFbov
control

Fig. 5

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SE 97/01344

A. CLASSIFICATION OF SUBJECT MATTER

IPC6: A61K 38/40, A23J 1/20
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC6: A61K, C07K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

SE,DK,FI,NO classes as above

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

WPI, CA, US PATENTS FULLTEXT, MEDLINE, BIOSIS, EMBASE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X ✓	EP 0506651 A2 (IMMUNO AKTIENGESELLSCHAFT), 30 Sept 1992 (30.09.92), claims 1-5 --	1-21
X ✓	EP 0568200 A2 (IMMUNO JAPAN INC.), 3 November 1993 (03.11.93), the abstract; page 4, lines 25 - 28; the claims --	1-10,12-21
X ✓	EP 0629347 A1 (MORINAGA MILK INDUSTRY CO., LTD.), 21 December 1994 (21.12.94), see page 3, lines 10 - 15; page 6, line 30 and the claims --	1-10,12-21

[X] Further documents are listed in the continuation of Box C. [X] See patent family annex.

* Special categories of cited documents:
"A" document defining the general state of the art which is not considered to be of particular relevance
"E" erlier document but published on or after the international filing date
"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
"O" document referring to an oral disclosure, use, exhibition or other means
"P" document published prior to the international filing date but later than the priority date claimed
"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"X" document of particular relevance: the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"Y" document of particular relevance: the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
12 November 1997	24 -11- 1997
Name and mailing address of the ISA/ Swedish Patent Office Box 5055, S-102 42 STOCKHOLM Facsimile No. +46 8 666 02 86	Authorized officer Carolina Palmcrantz Telephone No. +46 8 782 25 00

Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SE 97/01344

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	Chemical Abstracts, Volume 124, No 24, 10 June 1996 (10.06.96), (Columbus, Ohio, USA), page 1, THE ABSTRACT No 325373, JP,94-191487 A,, (Shimamura Seiichi et al) 15 August 1994 (15.08.94)	1-10,12-21
X	Dialog Information Service, file 155, Medline, Dialog accession no. 07325914, Medline accession no. 93146928, Bellamy W. et al: "Antibacterial spectrum of lactoferricin B, a potent bactericidal peptide derived from the N-terminal region of bovine lactoferrin", J Appl Bacteriol (ENGLAND) Dec 1992, 73 (6) P472-9	1-10,12-21
X	File WPI, Derwent accession no. 96-350155, Kuriiwa N: "Drug for inhibition of endotoxin-induced inflammation comprises peptide of mol. wt. 10000, derived from N-terminal region of comprises peptide of mol. wt. 10000, derived from N-terminal region of lactoferrin", JP,A,8165248, 960625, DW9635	1-9,11-21
X	Patent Abstracts of Japan, vol. 88, no.27, & JP,A,63051337 (Snow Brand Milk Prod Co Ltd) 4 March 1986	1-9,12-21
X	Patent Abstracts of Japan, vol. 96, no. 29, & JP,A,7309771 (Morinaga Milk Ind Co Ltd) 28 November 1995	1-9,12-21
P,X	EP 0730868 A1 (SATOH, TAMOTSU), 11 Sept 1996 (11.09.96), the claims	1-9,11-21
P,X	EP 0753308 A2 (GAMBIT INTERNATIONAL LIMITED), 15 January 1997 (15.01.97), see page 2, line 18 and the claims	1-10,12-21

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SE 97/01344

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
P,X	WO 9705884 A1 (NEW ENGLAND MEDICAL CENTER HOSPITALS, INC.), 20 February 1997 (20.02.97), the claims -- ---------	1-9,12-21

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SE 97/01344

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.: 15-21
because they relate to subject matter not required to be searched by this Authority, namely:

Remark: Although claims 15-21 are directed to a method of treatment of the human/animal body the search has been carried out and based on the alleged effects of the composition (c.f. PCT Rule 39.1(iv)).

2. [ ] Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. [ ] As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. [ ] As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest.

[ ] No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

Information on patent family members

01/10/97

International application No.
PCT/SE 97/01344

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
EP 0506651 A2	30/09/92	SE 0506651 T3	
		AT 65191 A	15/01/97
		AT 144148 T	15/11/96
		AT 402789 B	25/08/97
		CA 2063872 A,C	26/09/92
		DE 59207358 D	00/00/00
		HU 70212 A	28/09/95
		HU 9403398 D	00/00/00
		JP 5070370 A	23/03/93
EP 0568200 A1	03/11/93	CA 2093165 A	03/10/93
		JP 6145068 A	24/05/94
EP 0629347 A1	21/12/94	SE 0629347 T3	
		AU 665381 B	04/01/96
		DE 69220679 D	00/00/00
		US 5656591 A	12/08/97
		AU 2956492 A	01/09/93
		CA 2128612 A	05/08/93
		JP 5310594 A	22/11/93
		JP 5320067 A	03/12/93
		WO 9314640 A	05/08/93
JP 94-191487 A	15/08/94	NONE	
EP 0730868 A1	11/09/96	CA 2169810 A	18/08/96
		JP 8217693 A	27/08/96
EP 753308 A2	15/01/97	NONE	
WO 9705884 A1	20/02/97	AU 6641896 A	05/03/97